



US009234207B2

(12) **United States Patent**  
**Suelmann**

(10) **Patent No.:** **US 9,234,207 B2**  
(45) **Date of Patent:** **Jan. 12, 2016**

(54) **CUCUMBER VARIETY NUN 5545 CUP**

(71) Applicant: **Nunhems B.V.**, Nunhem (NL)

(72) Inventor: **Jos Suelmann**, Roermond (NL)

(73) Assignee: **Nunhems B.V.**, AC Nunhem (NL)

(\*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 439 days.

(21) Appl. No.: **13/668,817**

(22) Filed: **Nov. 5, 2012**

(65) **Prior Publication Data**

US 2013/0074203 A1 Mar. 21, 2013

(51) **Int. Cl.**

**A01H 5/08** (2006.01)

**C12N 15/82** (2006.01)

**A01C 14/00** (2006.01)

**A23L 1/212** (2006.01)

**A23K 1/14** (2006.01)

(52) **U.S. Cl.**

CPC ..... **C12N 15/8283** (2013.01); **A01C 14/00** (2013.01); **A01H 5/08** (2013.01); **A23K 1/14** (2013.01); **A23L 1/212** (2013.01); **A23L 1/2123** (2013.01); **C12N 15/8241** (2013.01); **C12N 15/8281** (2013.01)

(58) **Field of Classification Search**

None

See application file for complete search history.

(56) **References Cited**

**U.S. PATENT DOCUMENTS**

4,822,949 A 4/1989 Niego et al.  
5,349,128 A 9/1994 Quemada et al.

5,492,827 A 2/1996 Dirks  
6,084,152 A 7/2000 Kwak et al.  
6,765,130 B2 7/2004 Taurick

**OTHER PUBLICATIONS**

Eugenia Martin et al., Identification of markers linked to agronomic traits in globe artichoke, Australian Journal of Crop Science (2008) vol. 1(2), pp. 43-46.

Peter Vos et al., AFLP: a new technique for DNA fingerprinting 1995, Nucleic Acids Research (1995) vol. 23, No. 21, pp. 4407-4414.

A. B. Pisanu et al., Yield and Biometric Characteristics of 9 Clones Selected From the Population of "Spinoso Sardo" Artichokes, Acta Hort. (2004) vol. 660, pp. 83-89.

Colijn-Hooymans et al., Competence for regeneration of cucumber cotyledons is restricted to specific developmental stages, Plant Cell, Tissue and Organ Culture (1994) 39, pp. 211-217.

Sang-Gu et al., Callus growth and plant regeneration in diverse cultivars of cucumber (*Cucumis sativus* L.), Plant Cell, Tissue and Organ Culture (1988) 12, pp. 67-74.

D.A. Sarreb et al., Comparison of triploid and diploid cucumber in long-term liquid cultures, Plant Cell, Tissue and Organ Culture (2002) 71, pp. 231-235.

USDA Objective Description of Variety Cucumber (*Cucumis sativus* L.) as published by the US Department of Agriculture, <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELDEV3002687>.

UPOV descriptors, Guidelines for the Conduct of Tests for Distinctness, Uniformity and Stability, Cucumber, Gherkin, TG/61/7, Mar. 28, 2007, [http://www.upov.int/en/publications/tg-rom/tg061/tg\\_61\\_7.pdf](http://www.upov.int/en/publications/tg-rom/tg061/tg_61_7.pdf).

O'Sullivan, Processing Cucumber Variety Evaluation 2010.†

† cited by third party

*Primary Examiner* — Brent T Page

(57) **ABSTRACT**

The invention relates to the field of *Cucumis sativus*, in particular to a new variety of *Cucumis sativus* designated NUN 5545 CUP plants, seeds and cucumber fruits thereof.

**18 Claims, 2 Drawing Sheets**

Figure 1

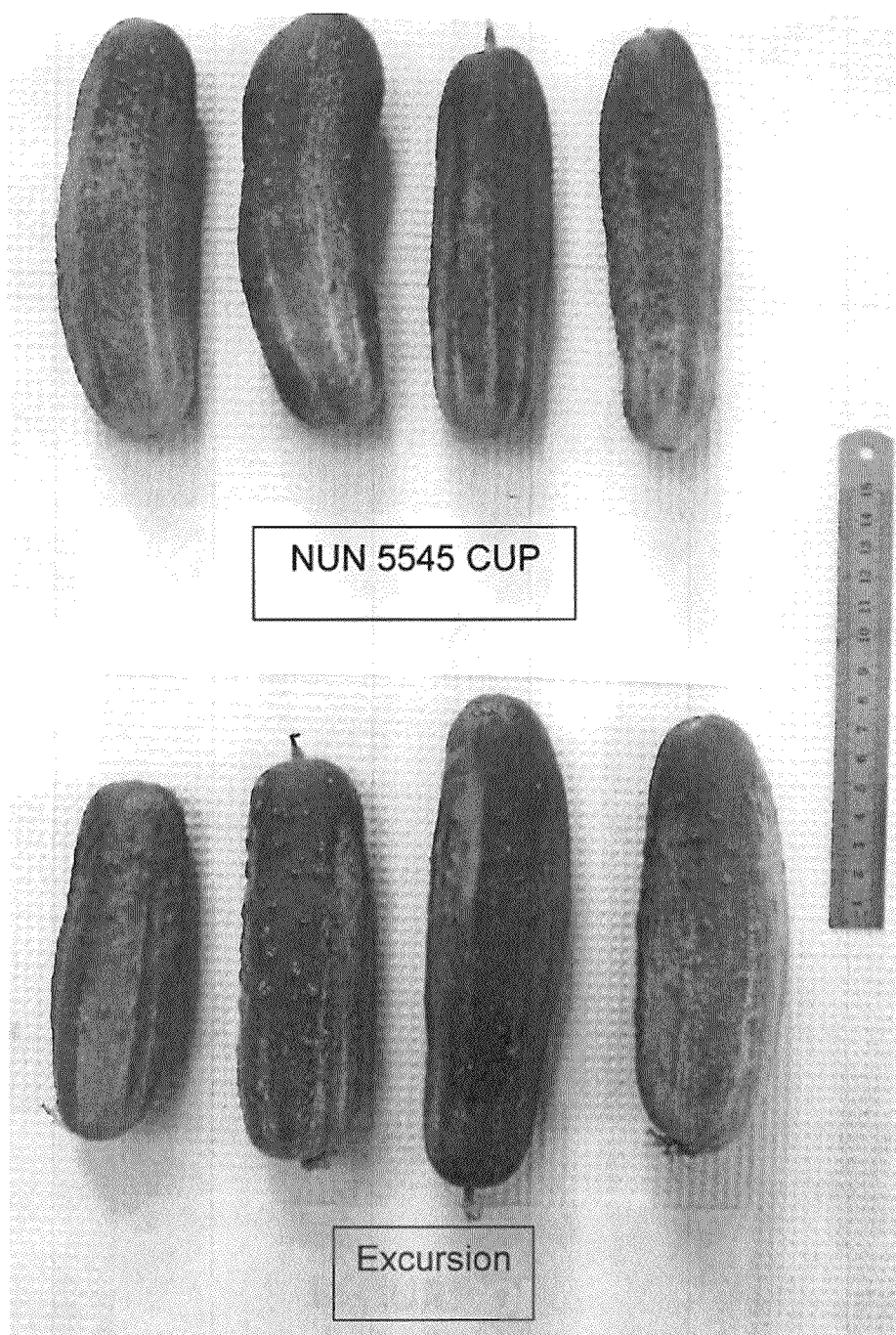


Figure 2



## CUCUMBER VARIETY NUN 5545 CUP

## FIELD OF THE INVENTION

The present invention relates to the field of plant breeding and, more specifically, to the development of cucumber variety NUN 5545 CUP.

The goal of vegetable breeding is to combine various desirable traits in a single variety/hybrid. Such desirable traits may include greater yield, resistance to insects or pests, tolerance to heat and drought, better agronomic quality, higher nutritional value, growth rate and fruit properties.

Breeding techniques take advantage of a plant's method of pollination. There are two general methods of pollination: a plant self-pollinates if pollen from one flower is transferred to the same or another flower of the same plant or plant variety. A plant cross-pollinates if pollen comes to it from a flower of a different plant variety.

Plants that have been self-pollinated and selected for type over many generations become homozygous at almost all gene loci and produce a uniform population of true breeding progeny, a homozygous plant. A cross between two such homozygous plants of different varieties produces a uniform population of hybrid plants that are heterozygous for many gene loci. Conversely, a cross of two plants each heterozygous at a number of loci produces a population of hybrid plants that differ genetically and are not uniform. The resulting non-uniformity makes performance unpredictable.

The development of uniform varieties requires the development of homozygous inbred plants, the crossing of these inbred plants, and the evaluation of the crosses. Pedigree breeding and recurrent selection are examples of breeding methods that have been used to develop inbred plants from breeding populations. Those breeding methods combine the genetic backgrounds from two or more plants or various other broad-based sources into breeding pools from which new lines are developed by selfing and selection of desired phenotypes. The new lines are evaluated to determine which of those have commercial potential.

One crop species which has been subject to such breeding programs and is of particular value is the cucumber. Cucumber (*Cucumis sativus* L.) is naturally a diploid ( $2n=14$ ) out-crossing species, although haploid, doubled-haploid (see, e.g., U.S. Pat. No. 5,492,827), and triploid (see, e.g., Sarreb et al. (2002), Plant Cell Tissue, Organ Culture 71: 231-235) types have been developed. The two main types of cucumber fruit grown commercially today in the United States are fresh market (slicing) type and the processing (pickling) type. Varieties and production methods are typically adapted to the end use. Slicing cucumbers are often longer, larger and have darker and thicker skin, whereas pickling/processing cucumbers have a shorter fruit, thinner skin with interior flesh that make them more amenable to pickling. Seedless varieties are generally preferable for both fresh market and for pickling as developing and large seeds are not palatable.

Until the 1960s cucumbers were normally monoecious, e.g., having separate male and female flowers on the same plant. Perfect flowers are uncommon in cucumbers. Staminate flowers are typically single and/or in clusters. Pistillate flowers may be solitary or in clusters and are borne on stout peduncles. Gynoecious cucumber plants have now been identified in which flowers are exclusively pistillate. These plants are generally higher yielding, due at least in part to the presence of higher numbers of female flowers. However, growth of gynoecious hybrid plants in the field has historically required the addition of plants of a monoecious line or variety (10-15%) to ensure availability of pollen and setting of fruit

with seed. Honey bees are the most commonly used insects to pollinate cucumbers in the open field.

Cucumber plants that set fruit parthenocarpically (without pollination and fertilization) have more recently been available. These plants produce seedless fruit unless pollinated. Growth of parthenocarpic varieties is beneficial in that setting of fruit on these cultivars does not produce an inhibiting effect on plant growth, unlike the case of fertilized, seeded fruit. The seedless varieties are usually higher yielding and of higher quality due to the lack of seeds. However, growth of these plants requires isolation from seeded cucumbers to avoid pollination and subsequent seeded fruit.

Most of the cucumbers currently used which are processed to pickles and pickle products in the United States are seeded hybrid varieties. Hybrid varieties offer the advantages of easy combination of dominant and recessive traits, such as disease resistance, from a set of inbred parents, as well as careful control of parentage. The production of F1 hybrid cucumber seeds from a pollen parent bearing only male flowers has been reported (see, e.g., U.S. Pat. No. 4,822,949).

Many different cucumber cultivars have been produced, and cucumber breeding efforts have been underway in many parts of the world (see e.g. U.S. Pat. No. 6,765,130). Some breeding objectives include varying the color, texture and flavor of the fruit. Minimizing the occurrence of bitterness in cucumbers is one such example. Other objectives include optimizing flesh thickness, solid content (% dry matter), and sugar content. Also, breeding programs have focused on developing plants with earlier fruit maturity, more restricted vine growth, improved disease resistance or tolerance, and improved adaptability to environmental conditions.

Advances in biotechnology have also resulted in genetically engineered cucumber plants with improved traits. For example, cucumbers resistant to CMV have been developed by expression of CMV protein coat genes (see e.g. U.S. Pat. No. 5,349,128). Transgenic plants exhibiting, for example, other viral resistance traits or high levels of superoxide dismutase have also been reported (see e.g. U.S. Pat. No. 6,084,152).

While breeding efforts to date have provided a number of useful cucumber varieties with beneficial traits, there remains a great need in the art for new varieties with further improved traits. Such plants would benefit farmers and consumers alike by improving crop yields and/or quality.

## SUMMARY OF THE INVENTION

In one aspect of the invention, a seed of cucumber variety NUN 5545 CUP is provided, wherein a representative sample of said seed has been deposited under Accession Number NCIMB 42404.

In another aspect the invention provides for a hybrid variety of *Cucumis sativus* called NUN 5545 CUP. The invention also provides for a plurality of seeds of the new variety, plants produced from growing the seeds of the new variety NUN 5545 CUP, and progeny of any of these. Especially, progeny retaining one or more (or all) of the "distinguishing characteristics" or one or more (or all) of the "essential morphological and physiological characteristics" or essentially all physiological and morphological characteristics of NUN 5545 CUP referred to herein, are encompassed herein as well as methods for producing these.

In one aspect, such progeny have (essentially) all the physiological and morphological characteristics of cucumber variety NUN 5545 CUP when grown under the same environmental conditions.

Further, a cucumber fruit produced on a plant grown from these seeds is provided.

In yet another embodiment of the invention, an Essentially Derived Variety of NUN 5545 CUP having one, two or three physiological and/or morphological characteristics which are different from those of NUN 5545 CUP and which otherwise has all the physiological and morphological characteristics of NUN 5545 CUP, wherein a representative sample of seed of variety NUN 5545 CUP has been deposited under Accession Number NCIMB 42404, is provided.

Further, a vegetatively propagated plant of variety NUN 5545 CUP, or a part thereof, is provided having all the morphological and physiological characteristics of NUN 5545 CUP when grown under the same environmental conditions.

Also a plant part derived from variety NUN 5545 CLIP is provided, wherein said plant part is selected from the group consisting of: harvested fruits or parts thereof, pollen, ovules, cells, leaves or parts thereof, petioles, shoots or parts thereof, stems or parts thereof, roots or parts thereof, cuttings, seeds, hypocotyl, cotyledon, flowers or parts thereof.

#### DEFINITIONS

“Cucumber” refers herein to plants of the species *Cucumis sativus*.

“Cultivated cucumber” refers to plants of *Cucumis sativus* i.e. varieties, breeding lines or cultivars of the species *C. sativus*, cultivated by humans and having good agronomic characteristics; preferably such plants are not “wild plants”, i.e. plants which generally have much poorer yields and poorer agronomic characteristics than cultivated plants and e.g. grow naturally in wild populations. “Wild plants” include for example ecotypes, PI (Plant Introduction) lines, landraces or wild accessions or wild relatives of a species.

The terms “cucumber plant designated NUN 5545”, “NUN 5545”, “NUN 5545 CUP” or “variety designated NUN 5545” are used interchangeably herein and refer to a cucumber plant of cucumber variety NUN 5545 CUP, representative seed of which having been deposited under Accession Number NCIMB 42404.

As used herein, the term “plant” includes the whole plant or any parts such as plant organs, plant cells, plant protoplasts, plant cell cultures or tissue cultures from which whole plants can be regenerated, plant callus, plant cell clumps, plant transplants, seedlings, plant cells that are intact in plants, plant clones or micropropagations, or parts of plants (e.g., harvested tissues, fruits or organs), such as plant cuttings, vegetative propagations, embryos, pollen, ovules, flowers, leaves, fruits, fruit flesh, seeds, clonally propagated plants, roots, stems, stalks, root tips, grafts, parts of any of these and the like, or derivatives thereof, preferably having the same genetic make-up for very similar genetic make-up) as the plant from which it is obtained. Also any developmental stage is included, such as seedlings, cuttings prior or after rooting, mature and/or immature plants or mature and/or immature leaves.

“Tissue culture” refers to a composition comprising isolated cells of the same or a different type or a collection of such cells organized into parts of a plant. Tissue culture of various tissues of cucumber and regeneration of plants therefrom is well known and widely published (see, e.g., Sang-Gu et al. (1988), Plant Cell, Tissue and Organ Culture 12: 67-74; Colijn-Hooymans (1994), Plant Cell, Tissue and Organ Culture 39: 211-217). Similarly, the skilled person is well-aware how to prepare a “cell culture”.

“UPOV descriptors” are the plant variety descriptors described for cucumber in the “Guidelines for the Conduct of

Tests for Distinctness, Uniformity and Stability, TG/61/7 (Geneva 2007), as published by UPOV (International Union for the Protection of New Varieties and Plants, available on the world wide web at upov.int) and which can be downloaded from the world wide web at [www.upov.int/en/publications/tg-rom/tg061/tg\\_61\\_7.pdf](http://www.upov.int/en/publications/tg-rom/tg061/tg_61_7.pdf) and is herein incorporated by reference in its entirety.

“USDA descriptors” are the plant variety descriptors for cucumber (*Cucumis sativus* L.) as published by the US Department of Agriculture, Agricultural Marketing Service, Science and Technology, Plant Variety Protection Office, Beltsville, Md. 20705 (available on the world wide web at [www.ams.usda.gov/AMSV1.0/](http://www.ams.usda.gov/AMSV1.0/)) and which can be downloaded from the world wide web at <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELDEV3002687>.

“RHS” refers to the Royal Horticultural Society of England which publishes an official botanical color chart quantitatively identifying colors according to a defined numbering system. The chart may be purchased from Royal Horticulture Society Enterprise Ltd RHS Garden: Wisley, Woking; Surrey GU236QB, UK, e.g., the RHS colour chart: 2007 (The Royal Horticultural Society, charity No: 222879, PO Box 313 London SW1P2PE; sold by, e.g., TORO-VERLAG, Obere Gruen 8 D-97877 Wertheim, Article-No.: Art62-00008 EAN-Nr.: 4250193402112). As used herein, the term “plant” includes the whole plant or any parts or derivatives thereof, preferably having the same genetic makeup as the plant from which it is obtained, such as plant organs (e.g. harvested or non-harvested fruits), plant cells, plant protoplasts, plant cell tissue cultures from which whole plants can be regenerated, plant calli, plant cell clumps, plant transplants, seedlings, hypocotyl, cotyledon, plant cells that are intact in plants, plant clones or micropropagations, or parts of plants (e.g. harvested tissues or organs), such as plant cuttings, vegetative propagations, embryos, pollen, ovules, fruits, flowers, leaves, seeds, clonally propagated plants, roots, stems, root tips, grafts, parts of any of these and the like. Also any developmental stage is included, such as seedlings, cuttings prior or after rooting, mature plants or leaves.

“Harvested plant material” refers herein to plant parts (e.g. fruits detached from the whole plant) which have been collected for further storage and/or further use.

“Harvested seeds” refers to seeds harvested from a line or variety, e.g. produced after self-fertilization or cross-fertilization and collected.

“Internode” refers to a portion of a plant stem between nodes.

“Node” refers to the place on a plant stem where a leaf is attached.

A plant having “(essentially) all the physiological and morphological characteristics” means a plant having the physiological and morphological characteristics when grown under the same environmental conditions of the plant from which it was derived, e.g. the progenitor plant, the parent, the recurrent parent, the plant used for tissue- or cell culture, etc. In certain embodiments the plant has all the physiological and morphological characteristics, except for certain characteristics mentioned, e.g. the characteristic(s) derived from a converted or introduced gene or trait and/or except for the characteristics which differ in an EDV. A plant have one or more “essential physiological and/or morphological characteristics” or one or more “distinguishing characteristics” refers to a plant having (or retaining) one or more of the characteristics mentioned in Table 1 when grown under the same environmental conditions that distinguish NUN 5545 CUP from comparison varieties (such as variety Excursion), such as but not limited to length of main stem, number of nodes from

cotyledon leaves to node bearing the first pistillate flower, number and form of tubercles, length of fruit seed at harvest maturity, number of seeds per fruit.

The physiological and/or morphological characteristics mentioned above are commonly evaluated at significance levels of 1%, 5% or 10% significance level, when measured under the same environmental conditions. For example, a progeny plant of NUN 5545 CUP may have one or more (or all) of the essential physiological and/or morphological characteristics of NUN 5545 CUP listed in Table 1, as determined at the 5% significance level when grown under the same environmental conditions.

As used herein, the term “variety” or “cultivar” means a plant grouping within a single botanical taxon of the lowest known rank, which grouping, irrespective of whether the conditions for the grant of a breeder’s right are fully met, can be defined by the expression of the characteristics resulting from a given genotype or combination of genotypes, distinguished from any other plant grouping by the expression of at least one of the said characteristics and considered as a unit with regard to its suitability for being propagated unchanged.

A variety is referred to as an “Essentially Derived Variety” (EDV) i.e., shall be deemed to be essentially derived from another variety, the initial variety; when (i) it is predominantly derived from the initial variety, or from a variety that is itself predominantly derived from the initial variety, while retaining the expression of the essential characteristics that result from the genotype or combination of genotypes of the initial variety; (ii) it is clearly distinguishable from the initial variety; and (iii) except for the differences which result from the act of derivation, it conforms to the initial variety in the expression of the essential characteristics that result from the genotype or combination of genotypes of the initial variety. Thus, an EDV may be obtained for example by the selection of a natural or induced mutant, or of a somaclonal variant, the selection of a variant individual from plants of the initial variety, backcrossing, or transformation by genetic engineering.

“Plant line” is for example a breeding line which can be used to develop one or more varieties.

“Hybrid variety” or “F1 hybrid” refers to the seeds harvested from crossing two inbred (nearly homozygous) parental lines. For example, the female parent is pollinated with pollen of the male parent to produce hybrid (F1) seeds on the female parent.

“Regeneration” refers to the development of a plant from cell culture or tissue culture or vegetative propagation.

“Vegetative propagation”, “vegetative reproduction” or “clonal propagation” are used interchangeably herein and mean the method of taking part of a plant and allowing that plant part to form at least roots where plant part is, e.g., defined as or derived from (e.g. by cutting of) leaf, pollen, embryo, cotyledon, hypocotyl, cells, protoplasts, meristematic cell, root, root tip, pistil, anther, flower, shoot tip, shoot, stem, fruit, petiole, etc. When a whole plant is regenerated by vegetative propagation, it is also referred to as a vegetative propagation.

“Selfing” refers to self-pollination of a plant, i.e., the transfer of pollen from the anther to the stigma of the same plant, “Crossing” refers to the mating of two parent plants.

“Average” refers herein to the arithmetic mean.

“Locus” (plural loci) refers to the specific location of a gene or DNA sequence on a chromosome. A locus may confer a specific trait.

“Allele” refers to one or more alternative forms of a gene locus. All of these loci relate to one trait. Sometimes, different alleles can result in different observable phenotypic traits,

such as different pigmentation. However, many variations at the genetic level result in little or no observable variation. If a multicellular organism has two sets of chromosomes, i.e. diploid, these chromosomes are referred to as homologous chromosomes. Diploid organisms have one copy of each gene (and therefore one allele) on each chromosome. If both alleles are the same, they are homozygotes. If the alleles are different, they are heterozygotes.

“Genotype” refers to the genetic composition of a cell or organism.

“Maturity” refers to the fruit developmental stage when the fruit has fully developed (reached its final size), begins to ripen and undergoes ripening, during which fruits can be divided into 1, 2, 3 or more maturity stages. Thereafter, fruits become overripe. In particular embodiments “maturity” is defined as the mature stage of fruit development and optimal time for harvest. In one embodiment a “mature” cucumber is defined as having reached the stage of maturity which will insure the proper completion of the normal ripening process. In particular embodiments, fruit should be harvested at a maturity stage i.e. substantially near maximum sweetness and flavor intensity.

“Harvest maturity” is referred to as the stage at which a cucumber fruit is ripe or ready for harvest or the optimal time to harvest the fruit. In one embodiment, harvest maturity is the stage which allows proper completion of the normal ripening.

“Flavor” refers to the sensory impression of a food or other substance, especially a cucumber fruit or fruit part (fruit flesh) and is determined mainly by the chemical senses of taste and smell. Flavor is influenced by texture properties and by volatile and/or non-volatile chemical components (organic acids, lipids, carbohydrates, etc.).

The term “traditional breeding techniques” encompasses herein crossing, selfing, selection, double haploid production, embryo rescue, protoplast fusion, marker assisted selection, mutation breeding etc as known to the breeder (i.e. methods other than genetic modification/transformation/transgenic methods), by which, for example, a genetically heritable trait can be transferred from one cucumber line or variety to another.

“Backcrossing” is a traditional breeding technique used to introduce a trait into a plant line or variety. The plant containing the trait is called the donor plant and the plant into which the trait is transferred is called the recurrent parent. An initial cross is made between the donor parent and the recurrent parent to produce progeny plants. Progeny plants which have the trait are then crossed to the recurrent parent. After several generations of backcrossing and/or selfing the recurrent parent comprises the trait of the donor. The plant generated in this way may be referred to as a “single trait converted plant”. “Progeny” as used herein refers to plants derived from a plant designated NUN 5545 CUP. Progeny may be derived by regeneration of cell culture or tissue culture or parts of a plant designated NUN 5545 CUP or selfing of a plant designated NUN 5545 CUP or by producing seeds of a plant designated NUN 5545 CUP. In further embodiments, progeny may also encompass plants derived from crossing of at least one plant designated NUN 6545 CUP with another cucumber plant of the same or another variety or (breeding) line, or wild cucumber plants, backcrossing, inserting of a locus into a plant or mutation. A progeny is, e.g., a first generation progeny, i.e. the progeny is directly derived from, obtained from, obtainable from or derivable from the parent plant by, e.g., traditional breeding methods (selfing and/or crossing) or regeneration. However, the term “progeny” generally encompasses further generations such as second, third, fourth, fifth, sixth, seventh



or more generations, i.e., generations of plants which are derived from, obtained from, obtainable from or derivable from the former generation by, e.g., traditional breeding methods, regeneration or genetic transformation techniques. For example, a second generation progeny can be produced from a first generation progeny by any of the methods mentioned above.

The terms “gene converted” or “conversion plant” in this context refer to cucumber plants which are developed by backcrossing wherein essentially all of the desired morphological and physiological characteristics of parent are recovered in addition to the one or more genes transferred into the parent via the backcrossing technique or via genetic engineering. Likewise a “Single Locus Converted (Conversion) Plant” refers to plants which are developed by plant breeding techniques comprising or consisting of backcrossing, wherein essentially all of the desired morphological and physiological characteristics of a cucumber variety are recovered in addition to the characteristics of the single locus having been transferred into the variety via the backcrossing technique and/or by genetic transformation.

“Transgene” or “chimeric gene” refers to a genetic locus comprising a DNA sequence which has been introduced into the genome of a cucumber plant by transformation. A plant comprising a transgene stably integrated into its genome is referred to as “transgenic plant”.

The term “mean” refers to the arithmetic mean of several measurements. The skilled person understands that the appearance of a plant depends to some extent on the growing conditions of said plant. Thus, the skilled person will know typical growing conditions for cucumbers described herein. The mean, if not indicated otherwise within this application, refers to the arithmetic mean of measurements on at least 10 different, randomly selected plants of a variety or line.

#### DRAWINGS

The drawings described herein are for illustration purposes only and are not intended to limit the scope of the present teachings in any way.

FIG. 1 shows typical fruits of NUN 5545 CUP and of variety Excursion at edible maturity.

FIG. 2 shows typical fruit seeds of NUN 5545 CUP and of variety Excursion at harvest maturity.

#### DETAILED DESCRIPTION

The present invention relates to a *Cucumis sativus* variety, referred to as NUN 5545 CUP, which has a longer main stem, a higher number of nodes from cotyledon leaves to node bearing the first pistillate flower, a longer length of fruit seed at harvest maturity and a higher amount of seeds per fruit than check variety Excursion. Moreover, NUN 5545 CUP has fruits with many obscure tubercles at the surface (e.g., similar to variety Straight Eight). Also encompassed by the present invention are progeny or EDVs of NUN 5545 CUP and methods of producing plants in accordance with the present invention.

A cucumber plant of NUN 5545 CUP differs from comparison variety Excursion in one or more characteristics (referred herein to as “distinguishing characteristics” or “distinguishing morphological and/or physiological characteristics” (or essential physiological and/or morphological characteristics) selected from I) the length of the main stem of NUN 5545 CUP is at least 10%, more preferably 20% longer compared to the length of the main stem of Excursion when using the length of the main stem of Excursion as reference (e.g.,

159 cm (NUN 5545 CUP) vs. 117 cm (Excursion)), II) the number of nodes from cotyledon leaves to node bearing the first pistillate flower of plants of NUN 5545 CUP is at least 10%, more preferably 20% higher than the number of nodes from cotyledon leaves to node bearing the first pistillate flower of plants of Excursion when using the number of nodes from cotyledon leaves to node bearing the first pistillate flower of Excursion as reference (e.g. 2.1 (NUN 5545 CUP) vs. 1.5 (Excursion)), III) fruits at edible maturity of NUN 5545 CUP show many obscure tubercles at their surface while fruits at edible maturity of Excursion show few prominent tubercles at their surface, IV) the length of fruit seed at harvest maturity of NUN 5545 CUP is at least 10% longer compared to the length of fruit seed at harvest maturity of Excursion when using the length of fruit seed at harvest maturity of Excursion as reference (e.g., 21.6 cm (NUN 5545 CUP) vs. 16.8 cm (Excursion)), V) the number of seeds per fruit of NUN 5545 CUP is at least 10% more compared to the number of seeds per fruit of Excursion when using the number of seeds per fruit of Excursion as reference (e.g., 119.6 seeds/fruit (NUN 5545 CUP) vs. 100 seeds/fruit (Excursion)).

It is understood that “significant” differences refer to statistically significant differences, when comparing the characteristic between two plant lines or varieties when grown under the same conditions. Preferably at least about 10, 15, 20 or more plants per line or variety are grown under the same conditions and characteristics are measured on at least about 10, 15, 20 or more randomly selected plant or plant parts to obtain averages. Thus, physiological and morphological characteristics or traits are commonly evaluated at a significance level of 1%, 5% or 10%, when measured in plants grown under the same environmental conditions.

Thus, in one aspect, the invention provides seeds of the cucumber variety designated NUN 5545 CUP wherein a representative sample of seeds of said variety was deposited under the Budapest Treaty, with Accession number NCIMB 42404.

Seeds of NUN 5545 CUP are obtainable by crossing the male parent with the female parent and harvesting the seeds produced on the female parent. The resultant NUN 5545 CUP seeds can be grown to produce NUN 5545 CUP plants. In one embodiment a plurality of NUN 5545 CUP seeds are packaged into small and/or large containers (e.g., bags, cartons, cans, etc.). The seeds may be treated with various compounds, such as seed coatings or fungicides or insecticides.

Also provided are plants of cucumber variety NUN 5545 CUP, or a fruit or other plant part thereof, produced from seeds, wherein a representative sample of said seeds has been deposited under the Budapest Treaty, with Accession Number NCIMB 42404. Also included is a cell culture or tissue culture produced from such a plant or a plant regenerated from such a cell or tissue culture said plant expressing all the morphological and physiological characteristics of NUN 5545 CUP.

Plants of NUN 5545 CUP can be produced by seeding directly in the ground (e.g., field) or by germinating the seeds in controlled environment conditions (e.g., greenhouses) and then transplanting the seedlings into the field. For example by sowing the seed into prepared seed beds where they will remain for the entire production of the crop. Alternatively, the cucumber seed may be planted through a black plastic mulch. The dark plastic will absorb heat from the sun, warming the soil early. It will also help to conserve moisture during the growing season, controls weeds and makes harvesting easier and cleaner. See for example [www.anrcatalog.ucdavis.edu](http://www.anrcatalog.ucdavis.edu) for cultivation, harvesting, handling and postharvest methods commonly used.

In another aspect, the invention provides for a cucumber plant of cucumber variety NUN 5545 CUP, a representative sample of seed from said variety has been deposited under the Budapest Treaty, with Accession number NCIMB 42404.

In other aspects, the invention provides for a fruit of cucumber variety NUN 5545 CUP, or a plant part, such as pollen, flowers, shoots or cuttings of variety NUN 5545 CUP or parts thereof.

In still another aspect the invention provides a method of producing a cucumber plant, comprising crossing a plant of cucumber variety NUN 5545 CUP with a second cucumber plant one or more times, and selecting progeny from said crossing.

In yet another aspect the invention provides a method of producing a cucumber plant, comprising selfing a plant of cucumber variety NUN 5545 CUP one or more times, and selecting progeny from said selfing.

In other aspects, the invention provides for progeny of variety NUN 5545 CUP such as progeny obtained by further breeding NUN 5545 CUP. Further breeding NUN 5545 CUP includes selfing NUN 5545 CUP one or more times and/or cross-pollinating NUN 5545 CUP with another cucumber plant or variety one or more times. In particular, the invention provides for progeny that retain all the essential morphological and physiological characteristics of NUN 5545 CUP or that retain one or more of the distinguishing characteristics of the cucumber type described further above and when grown under the same environmental conditions. In another aspect, the invention provides for vegetative reproductions of the variety and essentially derived varieties (EDVs) of NUN 5545 CUP.

The morphological and/or physiological differences between plants according to the invention, i.e. NUN 5545 CUP or progeny thereof, or an EDV thereof, and other known varieties can easily be established by growing NUN 5545 CUP next to the other varieties (in the same field, under the same environmental conditions), preferably in several locations which are suitable for said cucumber cultivation, and measuring morphological and/or physiological characteristics of a number of plants (e.g., to calculate an average value and to determine the variation range/uniformity within the variety). For example, trials can be carried out in Acampo Calif., USA (N 38 degrees 07'261"/W 121 degrees 18' 807", USA, whereby e.g. type of cucumber, area of best adaption in USA, days from seeding to harvest, predominate usage and culture, plant habit, plant growth, plant sex, flower color, main stem length, internode length, stem form, leaf length, leaf width, petiole length, fruit length, fruit diameter, fruit weight, fruit neck shape, fruit tapering, skin color/thickness/ribs/toughness/luster, spine color/quality/density, tubercles, flavor, fruit tapering fruit shape, fruit surface, seeds, disease resistances, insect resistances can be measured and directly compared.

Morphological and physiological characteristics (and distinguishing characteristics) of NUN 5545 CUP, are provided in the Examples, in Table 1. Encompassed herein are also plants derivable from NUN 5545 CUP (e.g. by selfings and/or crossing and/or backcrossing with NUN 5545 CUP and/or progeny thereof) comprising all the physiological and morphological characteristics of NUN 5545 CUP listed in Table 1 as determined at the 5% significance level when grown under the same environmental conditions and/or comprising one or more (or all; or all except one, two or three) of the distinguishing characteristics as determined at the 5% significance level when grown under the same environmental conditions.

Also at-harvest and/or post-harvest characteristics of fruits can be compared, such as cold storage holding quality (browning), post-harvest rind firmness and/or flesh firmness, and juiciness can be measured using known methods.

Flesh firmness can for example be measured using a penetrometer, e.g. by inserting a probe into the fruit flesh and determining the insertion force, or other methods.

The morphological and/or physiological characteristics may vary somewhat with variation in the environment (such as temperature, light intensity, day length, humidity, soil, fertilizer use), which is why a comparison under the same environmental conditions is preferred. Colors can best be measured against. The Munsell Book of Color (Munsell Color Macbeth Division of Kollmorgen Instruments Corporation) or using the Royal Horticultural Society Chart (<http://www.rhs.org.uk/Plants/RHS-Publications/RHS-colour-charts>).

In a preferred embodiment, the invention provides for cucumber fruits of variety NUN 5545 CUP, or a part of the fruit. In another embodiment, the invention provides for a container comprising or consisting of a plurality of harvested cucumber fruits of NUN 5545 CUP, or progeny thereof, or a derived variety, such as an EDV.

In yet a further embodiment, the invention provides for a method of producing a new cucumber plant. The method comprises crossing a plant of the invention NUN 5545 CUP, or an EDV thereof, or a progeny plant thereof, either as male or as female parent, with a second cucumber plant (or a wild relative of cucumber) one or more times, and/or selfing a cucumber plant according to the invention i.e. NUN 5545 CUP, or an EDV thereof, or a progeny plant thereof, one or more times, and selecting progeny from said crossing and/or selfing. The second cucumber plant may for example be a line or variety of the species *C. sativus* L., *Cucumis hystris*, *Cucumis ritchiei* (syn. *Dicaelospermum ritchiei*) or *Cucumis maderaspatana* (syn. *Mukia maderaspatana*).

Progeny are either the generation (seeds) produced from the first cross (F1) or selfing (S1), or any further generation produced by crossing and/or selfing (F2, F3, etc.) and/or backcrossing (BC1, BC2, etc.) one or more selected plants of the F1 and/or S1 and/or BC1 generation (or plants of any further generation, e.g. the F2) with another cucumber plant (and/or with a wild relative of cucumber). Progeny may have all the physiological and morphological characteristics of cucumber variety NUN 5545 CUP when grown under the same environmental conditions and/or progeny may have (be selected for having) one or more of the distinguishing characteristics of cucumbers of the invention. Using common breeding methods such as backcrossing or recurrent selection, one or more specific characteristics may be introduced into NUN 5545 CUP, to provide an EDV of NUN 5545 CUP.

The invention provides for methods of producing plants which retain all the morphological and physiological characteristics of NUN 5545 CUP. The invention provides also for methods of producing EDVs (Essentially Derived Varieties) of NUN 5545 CUP which differ from NUN 5545 CUP in one, two, three or more morphological and/or physiological characteristics, but which are still genetically closely related to NUN 5545 CUP. The relatedness can, for example be determined by fingerprinting techniques (e.g., making use of isozyme markers and/or molecular markers such as SNP markers, AFLP markers, microsatellites, minisatellites, RAPD markers, RFLP markers and others). A plant is "closely related" to NUN 5545 CUP if its DNA fingerprint is at least 80%, 90%, 95% or 98% identical to the fingerprint of NUN 5545 CUP. In a preferred embodiment AFLP markers are used for DNA fingerprinting (Vos et al. 1995, Nucleic



Acid Research 23: 4407-4414). A closely related plant may have a Jaccard's Similarity index of at least about 0.8, preferably at least about 0.9, 0.95, 0.98 or more (Pisanu et al. ISHS 2004, Acta Hort. 660). The invention also provides plants and varieties obtained by these methods. EDVs may be produced by crossing and/or selfing, or alternatively, an EDV may simply be identified and selected amongst NUN 5545 CUP plants, or progeny thereof, e.g. by identifying a variant within NUN 5545 CUP or progeny thereof (e.g. produced by selfing) which variant differs from NUN5545 CUP in one, two or three of the morphological and/or physiological characteristics (e.g. in one, two or three distinguishing characteristics), e.g. those listed in Table 1 or others.

By crossing and/or selfing also (one or more) single traits may be introduced into the variety of the invention i.e. NUN 5545 CUP (e.g., using backcrossing breeding schemes), while retaining the remaining morphological and physiological characteristics of NUN 5545 CUP and/or while retaining one or more distinguishing characteristics. A single trait converted plant may thereby be produced. For example, disease resistance genes may be introduced, genes responsible for one or more quality traits, yield, etc. Both single genes (dominant or recessive) and one or more QTLs (quantitative trait loci) may be transferred into NUN 5545 CUP by breeding with NUN 5545 CUP.

Any pest or disease resistance genes may be introduced into a plant according to the invention, i.e. NUN 5545 CUP, progeny thereof or into an EDV of NUN 5545 CUP. Resistance to one or more of the following diseases is preferably introduced into plants of the invention: Angular Leaf Spot (*Pseudomonas lachrymans*), Anthracnose (Race 1), *Colletotrichum lagenaria*, Anthracnose (Race 2), Bacterial Wilt (*Erwinia tracheiphilus*), Cucumber Scab (*Gummy*), *Cladosporium cucumerinum*, Downy Mildew, Powdery Mildew (*Erysiphe chicoracearum*), *Alternaria* Leaf Blight (*Alternaria cucumerina*), Target Spot (*Corynespora cassiicola*). Cucumber Yellow Mottle Mosaic Virus (*Cucumis* Virus 1), Cucumber Green Mottle Mosaic Virus (*Cucumis* Virus 2), Cucumber Aucuba Mosaic Virus (*Cucumis* Virus 2A), Muskmelon Mosaic Virus, Watermelon Mosaic Virus, Papaya Ring Spot Virus, Zucchini Mosaic Virus, Cucumber Rust, Root Rot, Crown Blight, Verticillium Wilt, Sulphur Burn, *Fusarium oxysporum* f.sp. *cucumeris* (Fom) race 0, *Fusarium oxysporum* f.sp. *cucumeris* (Fom) race 1, *Fusarium oxysporum* f.sp. *cucumeris* (Fern) race 2, *Fusarium* Wilt R2, Root Knot (Nematode), Anthracnose, and Squash Mosaic.

Resistance to one or more of the following pests is preferably present or introduced into plants of the invention: Aphid resistance, Pickle Worm, Darkling Ground Beetle, Banded Cucumber Beetle, Mite, Western Spotted Cucumber Beetle, Leafhopper, Cucumber Worm, Western Striped Cucumber Beetle or Leafminer. Other resistance genes, against pathogenic viruses, fungi, bacteria or pests may also be introduced.

Thus, invention also provides a method for developing a cucumber plant in a cucumber breeding program, using a cucumber plant of the invention, or its parts as a source of plant breeding material. Suitable plant breeding techniques are recurrent selection, backcrossing, pedigree breeding, mass selection, mutation breeding and/or genetic marker enhanced selection. For example, in one aspect, the method comprises crossing NUN 5545 CUP or progeny thereof, or an EDV thereof, with a different cucumber plant, and wherein one or more offspring of the crossing are subject to one or more plant breeding techniques selected from the group consisting of recurrent selection, backcrossing, pedigree breeding, mass selection, mutation breeding and genetic marker enhanced selection (see e.g. Martin et al. 2008, Australian

Journal of Crop Science 1(2): 43-46). For breeding methods in general see Principles of Plant Genetics and Breeding, 2007, George Acquaah, Blackwell Publishing, ISBN-13: 978-1-4051-3646-4.

The invention thus also provides a method of introducing a single locus conversion, or single trait conversion, into a cucumber plant according to the invention and/or into NUN 5545 CUP comprising:

- (a) crossing a cucumber plant of variety NUN 5545 CUP, a representative sample of seed of said variety having been deposited under Accession Number NCIMB 42404, with a second cucumber plant comprising a desired single locus to produce F1 progeny plants;
- (b) selecting F1 progeny plants that have the single locus to produce selected F1 progeny plants;
- (c) crossing the selected progeny plants with a plant of NUN 5545 CUP, to produce backcross progeny plants;
- (d) selecting backcross progeny plants that have the single locus and one or more (or all) distinguishing characteristics of cucumbers according to the invention and/or all the physiological and morphological characteristics of NUN 5545 CUP to produce selected backcross progeny plants; and
- (e) optionally repeating steps (c) and (d) one or more times in succession to produce selected second, third or fourth or higher backcross progeny plants that comprise the single locus and otherwise one or more (or all) the distinguishing characteristics of the cucumbers according to the invention and/or comprise all of the physiological and morphological characteristics of NUN 5545 CUP, when grown in the same environmental conditions.

The above method is provided, wherein the single locus confers a trait, wherein the trait is pest resistance or disease resistance.

In one embodiment the trait is disease resistance and the resistance is conferred to Angular Leaf Spot (*Pseudomonas lachrymans*), Anthracnose (Race 1), *Colletotrichum lagenaria*, Anthracnose (Race 2), Bacterial Wilt (*Erwinia tracheiphilus*), Cucumber Scab (*Gummy*), *Cladosporium cucumerinum*, Downy Mildew, Powdery Mildew (*Erysiphe chicoracearum*), *Alternaria* Leaf Blight (*Alternaria cucumerina*), Target Spot (*Corynespora cassiicola*). Cucumber Yellow Mottle Mosaic Virus (*Cucumis* Virus 1), Cucumber Green Mottle Mosaic Virus (*Cucumis* Virus 2), Cucumber Aucuba Mosaic Virus (*Cucumis* Virus 2A), Muskmelon Mosaic Virus, Watermelon Mosaic Virus, Papaya Ring Spot Virus, Zucchini Mosaic Virus, Cucumber Rust, Root Rot, Crown Blight, Verticillium Wilt, Sulphur Burn, *Fusarium oxysporum* f.sp. *cucumeris* (Fom) race 0, *Fusarium oxysporum* f.sp. *cucumeris* (Fom) race 1, *Fusarium oxysporum* f.sp. *cucumeris* (Fom) race 2, *Fusarium* Wilt R2, Root Knot (Nematode), Anthracnose, and Squash Mosaic.

In one embodiment the trait is pest resistance and the resistance is conferred to Aphid, Pickle Worm, Darkling Ground Beetle, Banded Cucumber Beetle, Mite, Western Spotted Cucumber Beetle, Leafhopper, Cucumber Worm, Western Striped Cucumber Beetle or Leaf miner.

The invention also provides a cucumber plant comprising at least a first set of the chromosomes of cucumber variety NUN 5545 CUP, a sample of seed of said variety having been deposited under Accession Number NCIMB 42404; further comprising a single locus conversion, wherein said plant has essentially all of the morphological and physiological characteristics of the plant comprising at least a first set of the chromosomes of cucumber NUN 5545 CUP. In another embodiment, this single locus conversion confers a trait selected from the group consisting of male sterility, herbicide

tolerance, insect resistance, pest resistance, disease resistance, environmental stress tolerance, modified carbohydrate metabolism and modified protein metabolism.

In one embodiment, NUN 5545 CUP may also be mutated (by e.g. irradiation, chemical mutagenesis, heat treatment, etc.) and mutated seeds or plants may be selected in order to change one or more characteristics of NUN 5545 CUP. Also natural mutants or natural variants of NUN 5545 CUP may be identified and used in breeding. Methods such as TILLING and/or EcoTILLING may be applied to cucumber populations in order to identify mutants. Similarly, NUN 5545 CUP may be transformed and regenerated, whereby one or more chimeric genes are introduced into the variety or into an EDV thereof. Transformation can be carried out using standard methods, such as *Agrobacterium tumefaciens* mediated transformation or biolistics, followed by selection of the transformed cells and regeneration into plants. A desired trait (e.g. genes conferring pest or disease resistance, herbicide, fungicide or insecticide tolerance, etc.) can be introduced into NUN 5545 CUP, or progeny thereof, by transforming NUN 5545 CUP or progeny thereof with a transgene that confers the desired trait, wherein the transformed plant retains all the phenotypic and/or morphological and/or physiological characteristics of NUN 5545 CUP or the progeny thereof and contains the desired trait.

The invention also provides for progeny of cucumber variety NUN 5545 CUP obtained by further breeding with NUN 5545 CUP. In one aspect progeny are F1 progeny obtained by crossing NUN 5545 CUP with another plant or S1 progeny obtained by selfing NUN 5545 CUP. Also encompassed are F2 progeny obtained by selfing the F1 plants. "Further breeding" encompasses traditional breeding (e.g., selfing, crossing, backcrossing), marker assisted breeding, and/or mutation breeding. In one embodiment, the progeny have one or more (for all) of the distinguishing characteristics mentioned further above when grown under the same environmental conditions. In a further embodiment the progeny have all the physiological and morphological characteristics of variety NUN 5545 CUP when grown under the same environmental conditions. In another embodiment the progeny are EDVs and/or have one, two, or three distinct traits (qualitative or quantitative) introduced into NUN 5545 CUP, while retaining all the other physiological and morphological characteristics of variety NUN 5545 CUP when grown under the same environmental conditions.

An EDV is an Essentially Derived Variety of NUN 5545 CUP having one, two or three physiological and/or morphological characteristics which are different from those of NUN 5545 CUP and which otherwise has all the physiological and morphological characteristics of NUN 5545 CUP, wherein a representative sample of seed of variety NUN 5545 CUP has been deposited under Accession Number NCIMB 42404. In particular variants which differ from NUN 5545 CUP in one, two or three of the characteristics mentioned in Table 1 are encompassed.

In one aspect, the EDV differs from NUN 5545 CUP in one, two or three of the distinguishing morphological and/or physiological characteristics selected from: I) length of main stem, II) number of nodes from cotyledon leaves to node bearing the first pistillate flower, III) number and form of tubercles, IV) length of fruit seed at harvest maturity, V) number of seeds per fruit.

In another embodiment the EDV may differ from NUN 5545 CUP in one, two or three morphological or physiological characteristic other than the "distinguishing morphological and/or physiological characteristics" (or essential physiological and/or morphological characteristics) of NUN 5545

CUP selected from: i) maturity (see, e.g. table 1, NUN 5545 CUP needs less days from seeding to market maturity than Excursion), ii) internode length, iii) predominant color at stem end of fruits at edible maturity, iv) predominant color at blossom end of fruits at edible maturity, v) spine quality of fruit at edible maturity, vi) spine density of fruit at edible maturity.

Cucumbers according to the invention, such as the variety NUN 5545 CUP, or its progeny, or an EDV of NUN 5545 CUP, can also be reproduced using vegetative reproduction methods. Therefore, the invention provides for a method of producing plants, or a part thereof, of variety NUN 5545 CUP, comprising vegetative propagation of variety NUN 5545 CUP. Vegetative propagation comprises regenerating a whole plant from a part of variety NUN 5545 CUP (or from its progeny or from an EDV of NUN 5545 CUP), such as a cutting, a cell culture or a tissue culture.

The invention also provides for a vegetatively propagated plant of variety NUN 5545 CUP for from its progeny or from an EDV of NUN 5545 CUP), or a part thereof, having one or more distinguishing characteristics and/or all the morphological and physiological characteristics of NUN 5545 CUP (except for the characteristics differing in the EDV), when grown under the same environmental conditions.

Parts of NUN 5545 CUP (or of its progeny or of an EDV of NUN 5545 CUP) encompass any cells, tissues, organs obtainable from the seedlings or plants, such as but not limited to, cucumber fruits or parts thereof, cuttings, hypocotyl, cotyledon, pollen and the like. Such parts can be stored and/or processed further. Encompassed are therefore also food or feed products comprising one or more of such parts, such as canned cucumber fruit from NUN 5545 CUP or from progeny thereof, or from a derived variety, such as an EDV.

In one aspect haploid plants and/or double haploid plants of NUN 5545 CUP, or an EDV or progeny of any of these, are encompassed herein. Haploid and double haploid (OH) plants can, for example, be produced by cell or tissue culture and chromosome doubling agents and regeneration into a whole plant. For DH production chromosome doubling may be induced using known methods, such as colchicine treatment or the like.

Also provided are plant parts derived from variety NUN 5545 CUP (or from its progeny or from an EDV of NUN 5545 CUP), or from a vegetatively propagated plant of NUN 5545 CUP (or from its progeny or from an EDV of NUN 5545 CUP), being selected from the group consisting of: harvested fruits or parts thereof, pollen, cells, leaves or parts thereof, petioles, cotyledons, hypocotyls, shoots or parts thereof, stems or parts thereof, roots or parts thereof, cuttings, or flowers.

In one embodiment, the invention provides for extracts of a plant described herein and compositions comprising or consisting of such extracts. In a preferred embodiment, the extract consists of or comprises tissue of a plant described herein or is obtained from such tissue.

The invention also provides for a food or feed product comprising or consisting of a plant part described herein preferably a cucumber fruit or part thereof and/or an extract from a plant part described herein. The food or feed product may be fresh or processed, e.g., canned, steamed, boiled, fried, blanched and/or frozen, etc.

For example, containers such as cans boxes, crates, bags, cartons, Modified Atmosphere Packagings, films (e.g. biodegradable films), etc. comprising plant parts of plants (fresh and/or processed) described herein are also provided herein.

Marketable cucumber fruits are generally sorted by size and quality after harvest.

All documents (e.g., patent publications) are herein incorporated by reference in their entirety.

## CITED REFERENCES

- Acquaah, Principles of Plant Genetics and Breeding, 2007, Blackwell Publishing, ISBN-13: 978-1-4051-3646-4
- Colijn-Hooymans (1994), Plant Cell, Tissue and Organ Culture 39: 211-217
- <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELDEV3002687>
- <http://www.rhs.org.uk/Plants/RHS-Publications/RHS-colour-charts>
- [http://www.upov.int/en/publications/tg-rom/tg061/tg\\_61\\_7.pdf](http://www.upov.int/en/publications/tg-rom/tg061/tg_61_7.pdf)
- Martin et al. 2008, Australian Journal of Crop Science 1(2): 43-46
- Pisanu et al. ISHS 2004, Acta Hort. 660
- Sang-Gu et al. (1988), Plant Cell, Tissue and Organ Culture 12:67-74
- Sarreb et al, (2002), Plant Cell Tissue, Organ Culture 71 231-235
- U.S. Pat. No. 4,822,949
- U.S. Pat. No. 5,349,128
- U.S. Pat. No. 5,492,827
- U.S. Pat. No. 6,084,152
- U.S. Pat. No. 6,765,130
- Vos et al 1995, Nucleic Acid Research 23 4407-4414

## EXAMPLES

## Development of NUN 5545 CUP

The hybrid NUN 5545 CUP was developed from a male and female proprietary inbred line of Nunhems. The female and male parents were crossed to produce hybrid (F1) seeds of NUN 5545 CUP. The seeds of NUN 5545 CUP can be grown to produce hybrid plants and parts thereof (e.g. cucumber fruit). The hybrid NUN 5545 CUP can be propagated by seeds or vegetative.

The hybrid variety is uniform and genetically stable. This has been established through evaluation of horticultural characteristics. Several hybrid seed production events resulted in no observable deviation in genetic stability. Coupled with the confirmation of genetic stability of the female and male parents the Applicant concluded that NUN 5545 CUP is uniform and stable.

## DEPOSIT INFORMATION

A total of 2500 seeds of the hybrid variety NUN 5545 CUP were deposited according to the Budapest Treaty by Nunhems B.V. on May 1, 2015, at the NCIMB Ltd., Ferguson Building Craibstone Estate, Bucksburn, Aberdeen AB21 9YA, United Kingdom (NCIMB). The deposit has been assigned Accession Number NCIMB 42404. A deposit of NUN 5545 CUP and of the male and female parent line is also maintained at Nunhems B.V. Access to the deposit will be available during the pendency of this application to persons determined by the Director of the U.S. Patent Office to be entitled thereto upon request. Subject to 37 C.F.R. §1.808(b), all restrictions imposed by the depositor on the availability to the public of the deposited material will be irrevocably removed upon the granting of the patent. The deposit will be maintained for a period of 30 years, or 5 years after the most recent request, or for the enforceable life of the patent whichever is longer, and will be replaced if it ever becomes nonviable during that period. Applicant does not waive any rights granted under this patent on this application or under the Plant Variety Protection Act (7 USC 2321 et seq.).

“USDA descriptors” are the plant variety descriptors for cucumber (*Cucumis sativus* L.)—Exhibit C of the U.S. Department of Agriculture, Agricultural Marketing Service, Science and Technology, Plant Variety Protection Office, Beltsville, Md. 20705, which can be downloaded from <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELDEV3002687> and which is herein incorporated by reference in its entirety.

The comparison variety to NUN 5545 CUP is Excursion a commercial variety from Seminis. In Table 1 a comparison between NUN 5545 CUP and Excursion is shown based on a trial in the USA. Trial location: Acampo Calif. USA, (coordinates: 38°07'261"N, -121° 18'80547"W), USA 2012. Average temperatures for the day were 33.1° C. and 14.7° C. for the night. Planting date: Jun. 20, 2012.

Two replications of 50 plants each, from which 20 plants or plant parts were randomly selected to measure characteristics. In Table 1 the USDA descriptors of NUN 5545 CUP (this application) and reference Excursion (commercial variety) are summarized.

In Table 1 distinguishing characteristics between NUN 5545 CUP and Excursion are highlighted in bold.

TABLE 1

| Physiological and/or morphological characteristics                      | NUN 5545 | Comparison Variety “Excursion” |
|---|----------|--------------------------------|
| <b>1. TYPE</b>  |          |                                |
| Predominate Usage<br>(1 = slicing; 2 = pickling)                        | 2        | 2                              |
| Predominate Culture<br>(1 = outdoor; 2 = indoor)                        | 1        | 1                              |
| Area of best adaptation (USA)<br>(1 = north; 2 = south; 3 = most areas) | 3        | 3                              |
| <b>2. MATURITY</b>  |          |                                |
| Days From Seeding To Market   | 46       | 50                             |
| <b>3. PLANT</b>   |          |                                |
| Habit<br>(1 = bush; 2 = semi-bush; 3 = vine)                            | 3        | 3                              |
| Growth<br>(1 = determinate; 2 = indeterminate)                          | 2        | 2                              |

TABLE 1-continued

| Physiological and/or morphological characteristics  | NUN 5545            | Comparison Variety "Excursion" |
|---|---------------------|--------------------------------|
| Flower color<br>(1 = yellow; 2 = orange; 3 = green; 4 = other)  | 1                   | 1                              |
| Color Chart Name  | RHS                 | RHS                            |
| Color Chart Value   | 14A (yellow-orange) | 14B (yellow-orange)            |
| <b>4. MAIN STEM</b>   |                     |                                |
| Length in cm  | 159                 | 117                            |
| Number of nodes from cotyledone leaves to node bearing the first pistillate flower  | 2.1                 | 1.5                            |
| Internode length in cm  | 2.1                 | 2.5                            |
| Stem form (1 = grooved, ridged; 2 = smooth, round)  | 1                   | 1                              |
| <b>5. LEAF</b>  |                     |                                |
| Length in mm  | 124                 | 123                            |
| Width in mm   | 172.8               | 165.4                          |
| Petiole length in cm  | 2.3                 | 2.1                            |
| <b>6. FRUIT AT EDIBLE MATURITY</b>  |                     |                                |
| Length in cm  | 12.9                | 12.8                           |
| Diameter at medial in cm  | 4.0                 | 4.4                            |
| Weight in g   | 132                 | 138.3                          |
| Skin color<br>(1 = not mottled; 2 = mottled or speckled with yellow)  | 2                   | 2                              |
| Yellowish blossomed end stripes<br>(1 = absent; 2 = extend less than 1/3 of fruit length; 3 = extend more than 1/3 of fruit length) | 3                   | 3                              |
| Predominant color at stem end<br>(1 = white; 2 = light green; 3 = medium green; 4 = dark green)                                     | 4                   | 3                              |
| Color Chart Name  | RHS                 | RHS                            |
| Color Chart Value   | 139A (green)        | 137A (green)                   |
| Predominant color at blossom end<br>(1 = white; 2 = light green; 3 = medium green; 4 = dark green)                                  | 2                   | 2                              |
| Color Chart Name  | RHS                 | RHS                            |
| Color Chart Value   | 145C (green)        | 145A (yellow green)            |
| Fruit neck shape<br>(1 = not necked; 2 = necked)  | 1                   | 1                              |
| Fruit tapering<br>(1 = both ends tapered; 4 = ends blunt or rounded)  | 4                   | 4                              |
| Stem end cross section<br>(1 = circular; 2 = triangular; 3 = square)  | 2                   | 2                              |
| Medial cross section<br>(1 = circular; 2 = triangular; 3 = square)  | 2                   | 2                              |
| Blossom end cross section<br>(1 = circular; 2 = triangular; 3 = square)   | 2                   | 2                              |
| Skin Thickness<br>(1 = thick; 2 = thin)   | 2                   | 2                              |
| Skin Ribs<br>(1 = not ribbed; 2 = ribbed)   | 2                   | 2                              |
| Skin toughness<br>(1 = tough; 2 = tender)   | 2                   | 2                              |
| Skin luster<br>(1 = dull; 2 = glossy)   | 1                   | 1                              |
| Spine color<br>(1 = white; 2 = black)   | 1                   | 1                              |
| Spine quality<br>(1 = coarse; 2 = fine)   | 2                   | 1                              |
| Spine density<br>(1 = few; 2 = many)  | 2                   | 1                              |
| Tubercles (warts)<br>(1 = few, obscure; 2 = many, obscure; 3 = few, prominent; 4 = many, prominent)                                 | 2                   | 3                              |
| Flavor<br>(1 = bitterfree; 2 = bitter)  | 1                   | 1                              |
| <b>7. FRUIT SEED AT HARVEST MATURITY</b>  |                     |                                |
| Length in cm  | 21.6                | 16.8                           |
| Diameter at medial in cm  | 6.3                 | 6.4                            |
| Color (1 = white; 2 = cream; 3 = yellow; 4 = orange; 5 = brown; 6 = red)  | 3                   | 3                              |
| Color Chart Name  | RHS                 | RHS                            |
| Color Chart Value   | 4C (yellow)         | 4C (yellow)                    |

TABLE 1-continued

| Physiological and/or morphological characteristics                      | NUN 5545 | Comparison Variety "Excursion" |
|---|----------|--------------------------------|
| Color pattern (1 = not striped; 2 = striped)                            | 1        | 1                              |
| Surface (1 = smooth; 2 = rough)   | 1        | 1                              |
| Netting (1 = slight or none; 2 = heavy)                                 | 1        | 1                              |
| Fruit set (1 = parthenocarpically; 2 = normally with seeds)             | 2        | 2                              |
| 8. SEEDS  |          |                                |
| No. per fruit   | 119.6    | 100                            |
| Weight in g/1,000 seeds   | 35.3     | 36                             |
| 9. DISEASE RESISTANCE<br>(0 = untested; 1 = susceptible; 2 = resistant) |          |                                |
| Angular Leaf Spot ( <i>Pseudomonas lachrymans</i> )                     | 2        | 2                              |
| Anthrachnose (Race 1) ( <i>Colletotrichum lagenaria</i> )               | 0        | 0                              |
| Anthrachnose ( <i>Colletotrichum orbiculare</i> )                       | 2        | 2                              |
| Bacterial Wilt ( <i>Erwinia tracheiphilus</i> )                         | 0        | 0                              |
| Cucumber Scab (Gummosis; <i>C. cucumerinum</i> )                        | 2        | 2                              |
| Downy Mildew  | 0        | 0                              |
| Powdery Mildew ( <i>Podosphaera xanthii</i> )                           | 2        | 2                              |
| <i>Alternaria</i> Leaf Blight ( <i>Alternaria cucumerina</i> )          | 0        | 0                              |
| Target Spot ( <i>Corynespora cassicola</i> )                            | 0        | 0                              |
| Cucumber Yellow Mottle Mosaic Virus (Cucumis Virus 1)                   | 0        | 0                              |
| Cucumber Green Mottle Mosaic Virus (Cucumis Virus 2)                    | 0        | 0                              |
| Cucumber Mosaic Virus   | 2        | 2                              |
| Muskmelon Mosaic  | 0        | 0                              |
| Others:   | 0        | 0                              |
| 10. INSECT RESISTANCE<br>(0 = untested; 1 = susceptible; 2 = resistant) |          |                                |
| Aphid ( <i>Aphis gossypii</i> )   | 0        | 0                              |
| Pickleworm ( <i>Diaphania nitidalis</i> )                               | 0        | 0                              |
| Eastern Striped Cucumber Beetle ( <i>Acolymma vittata</i> )             | 0        | 0                              |
| Two Spotted Mite ( <i>Tetranychus bimaculatus</i> )                     | 0        | 0                              |
| Spotted Cucumber Beetle ( <i>Diabrotica undecimpunctata howardi</i> )   | 0        | 0                              |
| Western Striped Cucumber Beetle ( <i>Acolymma trivittata</i> )          | 0        | 0                              |
| Other (Specify)   | 0        | 0                              |

These are typical values. Values may vary due to environment. Other values that are substantially equivalent are also within the scope of the invention.

N.A. = not applicable;

n.r. = not recorded.

The invention claimed is:

1. A seed of cucumber variety NUN 5545 CUP, wherein a representative sample of said seed has been deposited under Accession Number NCIMB 42404.

2. A plant of cucumber variety NUN 5545 CUP, or a part thereof, wherein a representative sample of seed of said variety has been deposited under Accession Number NCIMB 42404.

3. A fruit of cucumber variety NUN 5545 CUP, or a plant part produced from the plant of claim 2.

4. A method of producing a cucumber plant, comprising crossing the plant of claim 2 with a second cucumber plant one or more times, and selecting progeny from said crossing.

5. A method of producing a cucumber plant, comprising selfing the plant of claim 2 one or more times, and selecting progeny from said selfing.

6. A Single Locus Converted Plant of NUN 5545 CUP having one physiological and/or morphological characteristic which is different from those of NUN 5545 CUP and which otherwise has all the physiological and morphological characteristics of NUN 5545 CUP, wherein a representative sample of seed of variety NUN 5545 CUP has been deposited under Accession Number NCIMB 42404.

7. A method of producing plants, or a part thereof, of variety NUN 5545 CUP comprising vegetative propagation of variety NUN 5545 CUP.

8. The method of claim 7, wherein said vegetative propagation comprises regenerating a whole plant from a part of variety NUN 5545 CUP.

9. The method of claim 8, wherein said part is a cutting, a cell culture or a tissue culture.

10. A vegetative propagated plant of variety NUN 5545 CUP, or a part thereof, having all the morphological and physiological characteristics of NUN 5545 CUP when grown under the same environmental conditions.

11. Plant part derived from variety NUN 5545 CUP, or from a plant of claim 10, wherein said plant part are harvested fruit or parts thereof, pollen, cells, leaves or parts thereof, petioles, shoots or parts thereof, stems or parts thereof, roots or parts thereof, cuttings, or flowers or parts thereof.

12. A food or feed product comprising a plant part of claim 11.

13. The food or feed product of claim 12, wherein said plant part is fresh or processed.

14. A cucumber plant produced by growing the seed of claim 1.

**21**

**15.** A method of producing a cucumber plant having a desired trait, wherein the method comprises transforming the cucumber plant of claim **2** with a transgene that confers the desired trait, wherein the transformed plant retains all the phenotypic and morphological characteristics of variety NUN 5545 CUP and contains the desired trait, a representative sample of seed of said variety NUN 5545 CUP having been deposited under Accession Number NCIMB 42404.

**16.** A cucumber plant produced by the method of claim **15**, wherein the plant comprises the desired trait and all of the physiological and morphological characteristics of NUN 5545 CUP.

**17.** A cell or tissue culture produced from a plant of claim **2**.

**18.** A cucumber plant regenerated from a cell or tissue culture of claim **17**, said plant expressing all the morphological and physiological characteristics of NUN 5545 CUP, wherein a representative sample having been deposited under Accession Number NCIMB 42404.

\* \* \* \* \*

20

**22**